1. **[TWICE AMENDED]** A method of detecting DNA variation by monitoring the formation or dissociation of a complex consisting of:

   (a) a single DNA strand of a double stranded DNA of at least 40 base pairs containing the locus of a variation bound to a solid surface,

   (b) an oligonucleotide or DNA analogue probe specific for one allele of the variation and capable of hybridizing to the single strand (a) to form a DNA duplex,

   (c) an intercalating dye specific for the DNA duplex structure of (a) plus (b) which forms a complex with the said duplex and reacts uniquely when interacting within the DNA duplex,

   which method comprises:

   (1) continually measuring an output signal indicative of interaction of the dye with duplex formed from the strand (a) and probe (b), and

   (2) recording the temperature at which a change in reaction output signal occurs which is attributable to formation or dissociation of the complex and is thereby correlated with the strength with which the probe (b) has hybridized to the single strand (a).

7. **[AMENDED]** A method according to claim 1, in which the single strand is bound to the solid surface by a biotin/streptavidin type interaction.

9. **[AMENDED]** A method according to claim 8, in which the buffer solution is Hepes buffer having a salt concentration less than 200 mM.

11. **[AMENDED]** A method according to claim 1, in which double stranded DNA is a product of PCR amplification of a target sequence.

14. **[TWICE AMENDED]** A method of detecting DNA variation which comprises bringing together:

(a) a single DNA strand of a double stranded DNA of at least 40 base pairs containing the locus of a variation bound to a solid surface,

(b) an oligonucleotide or DNA analogue probe specific for one allele of the variation and capable of hybridizing to the single strand (a) to form a DNA duplex,

(c) an intercalating dye specific for the DNA duplex structure of (a) plus (b) which forms a complex with the said duplex and reacts uniquely when interacting within the duplex,

thereby forming a complex consisting of the components (a), (b) and (c), wherein the components (a), (b), and (c) are brought together under conditions in which either

(i) the component (a) hybridizes to component (b) and the complex is formed with component (c), or

(ii) the components (a) and (b) do not hybridize and the complex with component (c) is not formed,

(2) thereafter steadily and progressively adjusting the temperature, respectively, either

(i) to denature the formed DNA duplex and cause dissociation of the complex, or

(ii) to cause formation of the DNA duplex and resulting complex,

(3) continually measuring an output signal indicative of the extent of hybridization of (a) and (b) and resulting complex formation with (c), and

(4) recording the temperature at which a change of output signal occurs which is indicative of, respectively,

(i) dissociation of the complex, or

(ii) formation of the complex.

20. **[AMENDED]** A method according to claim 14, in which the single strand is bound to the solid surface by a biotin/streptavidin type interaction.

22. **[AMENDED]** A method according to claim 21, in which the buffer solution is Hepes buffer having a salt concentration less than 200 mM.

24. **[AMENDED]** A method according to claim 14, in which the double stranded DNA is a product of PCR amplification of a target sequence.

27. **[TWICE AMENDED]** A method of detecting DNA variation which comprises:

(1) forming a complex consisting of:

(a) a single DNA strand of a double stranded DNA of at least 40 base pairs containing the locus of a variation bound to a solid surface,

(b) an oligonucleotide or DNA analogue probe specific for one allele of the variation hybridized to the single strand (a) to form a duplex, and

(c) an intercalating dye specific for the DNA duplex structure of (a) plus (b) and which reacts uniquely when interacting within the DNA duplex, and

(2) continually measuring an output signal of the extent of the resulting reaction of the marker and the duplex while steadily increasing the temperature,

(3) recording the temperature at which a change in reaction output signal occurs which is attributable to dissociation of the complex and is thereby correlated with the strength with which the probe (b) has hybridized to the single strand (a).

33. **[AMENDED]** A method according to claim 27, in which the single strand is bound to the solid surface by a biotin/streptavidin type interaction.

35. **[AMENDED]** A method according to claim 34, in which the buffer solution is Hepes buffer having a salt concentration less than 200 mM.

37. **[AMENDED]** A method according to claim 27, in which the double stranded DNA is a product of PCR amplification of a target sequence.

40. **[TWICE AMENDED]** A method of detecting DNA variation which comprises:

(1) bringing together:

(a) a single DNA strand of a double stranded DNA of at least 40 base pairs containing the locus of a variation bound to a solid surface,

(b) an oligonucleotide or DNA analogue probe specific for one allele of the variation and capable of hybridizing to the single strand (a) to form a DNA duplex,

(c) an intercalating dye specific for the DNA duplex structure of (a) plus (b) and which reacts uniquely when interacting within the duplex,

the components (a), (b) and (c) being brought together prior to formation of the defined complex and under conditions in which (a) and (b) do not hybridize;

(2) steadily adjusting the temperature to cause formation of the duplex and resulting complex consisting of components (a), (b), and (c), and

(3) measuring an output signal indicative of the occurrence of hybridization of (a) and (b) (herein termed the annealing point).

46. **[AMENDED]** A method according to claim 40, in which the single strand is bound to the solid surface by a biotin/streptavidin type interaction.

48. **[AMENDED]** A method according to claim 47, in which the buffer solution is Hepes buffer having a salt concentration less than 200 mM.

50. **[AMENDED]** A method according to claim 40, in which the double stranded DNA is a product of PCR amplification of a target sequence.